TABLE 1

SHOWING EXTRACT OF MATURE CORNSTALKS

Sample No.	Dry weight, % of fresh weight	Total water, g	Brix, corr. to 20° C	Extract, % of dry weight	Extract, % of stalks
1	22.0	297	2.11	21.3	4.69
2	26.53	326	4.28	48.6	12.86
3	18.68	294	3.57	35.9	6.71
4	15.36	297	2.38	24.1	3.70
5	32.77	295	2.08	20.9	6.85
6	28.21	330	3.28	37.3	11.52
7	23.36	306	3.68	39.0	10.52
8	24.06	305	3.03	31.8	7.65
9	32.83	305	4.88	52.1	17.10
10	28.18	303	3.98	39.9	11.24
11	26.67	319	2.78	36.1	9.63
12	18.38	302	2.78	28.8	5.29

thirds of the soluble solids of normal mature cornstalks consist of sugars.

The method was used here to measure the extract content of stalks of different varieties of hybrid corn that had borne mature ears.[1] Single stalks of 12 strains of hybrid corn were analyzed. The data, although not representing statistical averages, as would have been desirable, illustrate the application of the method and show in general the magnitude of the soluble solids differences found among stalks of different genetic origin.

Each stalk was finely ground by passing it through a motorized saw-blade grinder (*2*). The ground material was weighed, spread on a cloth-covered tray, and dried to constant weight at slightly below 50° C in an electrically heated dryer. It has been shown by Sayre and Morris (*4*) that drying cornstalk tissue at about 48° C gives correct dry weight percentages.

Thirty-gram samples of dry cornstalk were mixed in tared glass jars with approximately 300 ml of hot water, let stand to cool and to allow time for equilibrium to become established, then weighed, pressed in a tincture press, and the sp g of the juice measured by Brix spindle. To calculate percentage of extract, the weight of water present was divided by 100, minus the corrected Brix reading, and multiplied by 100. This gave weight of juice. This value, times the corrected Brix reading and divided by 100, gave the weight of juice solids in the 30 g of sample taken in terms of the sugar scale. Example: the corrected Brix was 2.11 and the total water present was 297 g, making the weight of juice 303.4 g. The weight of juice solids was $303.4 \times 2.11 \div 100$, or 6.40 g, or 21.3% of the sample taken.

The data are shown in Table 1.

Unexpectedly wide differences appeared in the extract percentages of the samples of different inheritance—20.9–52.1% on the dry matter basis, and 3.70–17.10% in terms of percentage of stalks. The high result of 52.1% is not new, as the value found by chemists of the Ohio Agricultural Experiment Station (*5*) in the case of the Burr-Leaming variety at maturity was a total sugar content of 35.6% on the dry matter basis, equal to 53.4% of soluble solids on the assumption that the sugars present were equal to two-thirds of the extract.

The extent to which these differences were due to genetic factors is unknown. If inheritance is indeed a controlling factor, an inexpensive method is evidently available for producing high sugar cornstalks: consisting of producing stalks from seed adapted to the locality, and having the property of producing stalks rich in extract at the time when the grain has reached maturity.

[1] Samples were obtained through the courtesy of J. D. Sayre, of the Ohio Agricultural Experiment Station, and were received in Scarsdale, New York, on September 28 by express from Wooster, Ohio. The stalks had been freed from ears, husks, leaves, leaf sheathes, and tassels and carefully packed in moistureproof bags. They were received in excellent condition.

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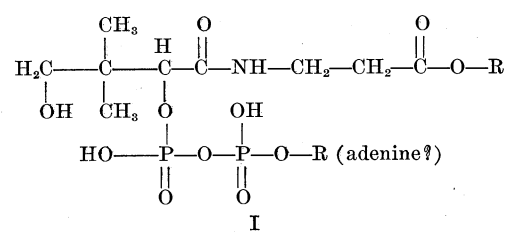
Phosphates of Pantothenic Acid[1, 2]

Tsoo E. King and F. M. Strong[3]

Department of Biochemistry, College of Agriculture
University of Wisconsin, Madison

In view of the fact that phosphate derivatives of most B vitamins, rather than the free forms, take part in active biochemical processes and of the fact that both fractions of coenzyme A contain phosphorus (*1*), efforts have been under way for some time in this laboratory to prepare various phosphates of pantothenic acid and to test their biological activities. Recently Lipmann, Novelli, and coworkers have suggested that degradation products encountered during the isolation of coenzyme A may be resynthesized into the coenzyme and hence show acetylation activity *in vitro* in a crude pigeon liver enzyme system. On the basis of this and other work, especially degradation by various enzymes, they have proposed for the coenzyme the partial structure I (*2*):



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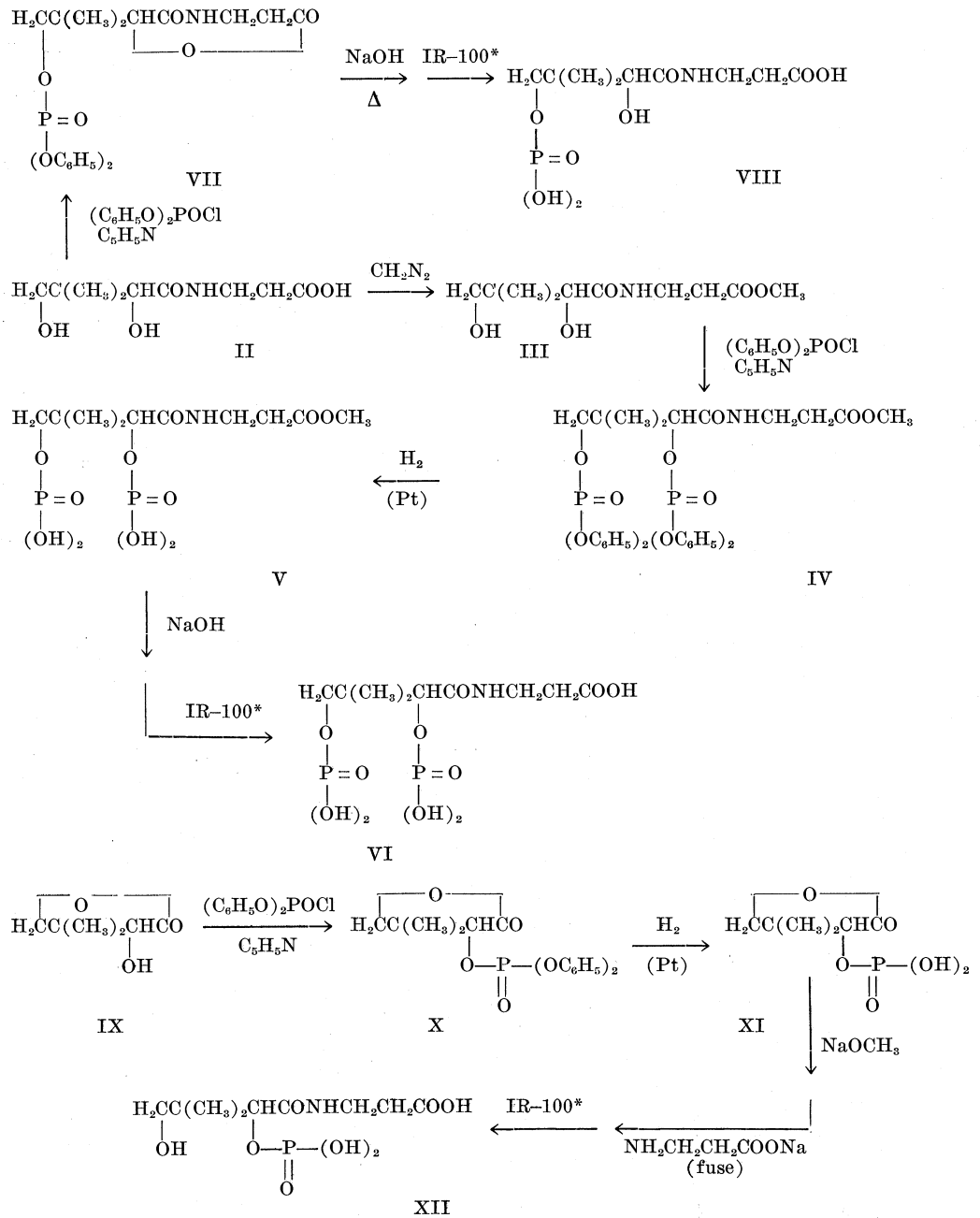
[2] This investigation was supported in part by research grants from the National Institutes of Health, USPH.

[3] The authors wish to acknowledge the assistance of Winifred Rogers and Harvey Higgins in carrying out tests for pantothenic acid and coenzyme A activity.



* IR–100 = Amberlite IR–100, H—form.

FIG. 1. Synthesis of pantothenic acid phosphates.

It therefore seems desirable to report at this time a preliminary account of our work on pantothenic acid phosphates.

Woolley in 1940 reported the preparation of pantothenic acid diphosphate by direct phosphorylation of the vitamin with phosphorus oxychloride (*3*). Unfortunately several investigators have been unable to obtain the diphosphate by his method (*4*, *5*, *6*). Accordingly, other phosphorylating agents were studied, and diphenylchlorophosphonate (*7*) was found satisfactory. The synthetic reactions carried out are summarized in Fig. 1. Methyl pantothenate (III), gave the bis-diphenylphosphate (IV) as expected, and removal of the phenyl groups from IV by reductive cleavage with hydrogen in the presence of Adam's catalyst gave methyl diphosphopantothenate, V, in good yield.

Anal. Calculated for $C_{10}H_{21}O_{11}NP_2 \cdot CH_3OH$: P, 14.59; N, 3.29.
Found: P (Fiske-SubbaRow), 14.2, N (Kjeldahl), 3.18.

Hydrolysis to the free acid, VI, was carried out in alkaline solution.

In contrast to the methyl ester, the reaction of free pantothenic acid with diphenylchlorophosphonate gave only a mono-diphenylphosphate derivative.

Anal. Calculated for $C_{21}H_{24}O_7NP$: C, 58.20; H, 5.57; N, 3.23; P, 7.15.
Found: C, 58.86; H, 6.95; N (Kjeldahl), 3.13; P (Fiske-SubbaRow), 6.8.

This result, and the fact that the product did not show any acidic characteristics, led us to formulate the product as the "gamma" phosphate, VII. This structure has not been otherwise established. The product, VII, was resistant to hydrogenolysis, but the phenyl groups were readily removed by alkaline hydrolysis to produce the free monophosphate, VIII. Although compounds VI and VIII have not been isolated in crystalline form as yet, the ratios of phosphorus content to pantothenic acid activity after enzymatic dephosphorylation closely approximate the theoretical values for the structures indicated.

The synthetic approach to the "alpha" monophosphate, XII, is outlined in Fig. 1. Pantoyl lactone was easily phosphorylated with diphenylchlorophosphonate as previously shown in this laboratory (*16*). Cleavage of the phenyl groups of the product was accomplished in this case more readily by hydrogenolysis than by alkaline hydrolysis. The free lactone phosphate, XI, was isolated as a crystalline product, mp 188°–189°.

Anal. Calculated for $C_6H_{11}O_6P$: P, 14.8.
Found: P (Fiske-SubbaRow), 14.3.

The coupling of the disodium salt of XI with sodium β-alanate was made by direct fusion. After removing the sodium by ion exchange a crude product was obtained which was strongly acidic, free from inorganic phosphate (less than 0.1%), and showed typical bound pantothenic acid activity (see below). Since the starting materials did not possess such activity, it seems quite certain that the "alpha" phosphate, XII, was present.

All the reactions illustrated in Fig. 1 gave over 70% yield except the coupling of phosphopantoyl lactone with β-alanine, and the alkaline hydrolysis of the "gamma" mono-diphenylphosphate, VII. All three preparations (VI, VIII, and XII) were free from inorganic phosphate (less than 0.1%). Aqueous solutions of the free acids were highly acidic, and of the normal sodium or potassium salts, alkaline. The barium and calcium salts were soluble in water.

The pantothenic acid phosphates, like the natural bound forms of pantothenic acid (e.g., coenzyme A [*8*] and PAC [*9*]), were inactive to *Lactobacillus arabinosus*. However, the pantothenic acid could be liberated through treatment with intestinal phosphatase (*1*) and was thereby rendered available to lactic acid bacteria (Table 1). Other enzymes, like papain, pancreatin, and mylase-P, were ineffective for the liberation. Phosphopantoyllactone (XI) was slightly active to *Acetobacter suboxydans*, but the phosphate derivatives of pantothenic acid were inert. Coenzyme A and PAC (*10, 11*) showed higher activity than the free vitamin in this organism. The present results demonstrate the high specificity of assimilation by the organism in that the free vitamin apparently combines with other components (possibly glutamic acid) and then is phosphorylated, whereas the prephosphorylated vitamins are not assimilable.

Both free pantothenic acid and methyl pantothenate are very sensitive to alkali. However, the vitamin activity of pantothenic acid phosphates was almost completely retained even after heating aqueous solutions in *N* sodium hydroxide in a boiling water bath for 1 hr (cf. Table 1).

TABLE 1

THE STABILITY OF PANTOTHENIC ACID PHOSPHATE

Compound tested	Treatment			
	None		N NaOH in boiling water bath for 1 hr	
	Free P.A.*	Total P.A.†	Free P.A.*	Total P.A.†
	Percentage of P.A. activity			
Calcium pantothenate	100	100	0	0
Methyl pantothenate	26	26	0	0
Pantothenic acid diphosphate (IV)	2	100	2	80–100

* P.A. = pantothenic acid.
† As measured with *L. arabinosus* after double enzyme treatment (*1*).

At the same time no inorganic phosphate was liberated. The natural existence of alkali-stable forms of pantothenic acid has been previously reported from this laboratory (*1, 12*). Pantothenic acid diphosphate was also more stable than the free vitamin to acid. After heating in 1 *N* hydrochloric acid solution for 1 hr in a boiling water bath, 30–70% of the bound pantothenic acid activity was still intact.

In the crude pigeon liver system (*13*) pantothenic acid diphosphate in the presence of adenosine triphosphate showed unmistakable coenzyme A activity in several trials, although the results were erratic and not consistently reproducible. This occasional activity was probably due to a synthesis of the coenzyme from the pantothenic acid diphosphate and other components of the test system. Similar examples of enzymatic synthesis of other B-vitamin-containing coenzymes have been reported (*14, 15*). Further investigations along this line, and on the biological activity of the pantothenic acid phosphates per se, are in progress.

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The Abundance of Several Relatively Rare Elements in Igneous Rocks of North America[1]

L. H. Ahrens and Lorraine G. Gorfinkle

Department of Geology, Massachusetts Institute of Technology, Cambridge

Methods of precision spectrochemical analysis, which have recently been developed in the Department of Geology, MIT, are being applied to the quantitative analysis of 17 relatively rare elements in approximately 150 igneous rock specimens from North America. The whole project will involve 2,000–2,500 determinations, each in duplicate. Thus far, about 1,500 determinations have been made. There will, however, be considerable delay before the project is complete, and all the data assembled and discussed; this note is a preliminary report.

These elements are being determined: Li, Rb, Cu, Ag, Ga, Pb, La, Y, Nd, Sc, V, Co, Cr, Sr, Ba, Zr, and F. The present investigation is concentrated mainly on some common rock types, namely, diabase, granite, and basalt; later investigations may include other rock types, including sediments. One object of the investigation is the determination of the abundance of the above elements in igneous rocks of North America, first, for the sake of intercontinental comparisons and, second, to check existing abundance values for these elements in the earth's crust and to provide new and more precise values. Another object is a statistical survey of the abundance distribution of each element within a given rock type. Because precision methods are being used and because many specimens of each rock type have been analyzed, the analytical data may be handled statistically on a quantitative basis. For example, it is a well-known *qualitative* fact that gallium is relatively uniformly distributed in igneous rocks because of its close association with aluminum. Fig. 1 is a histogram

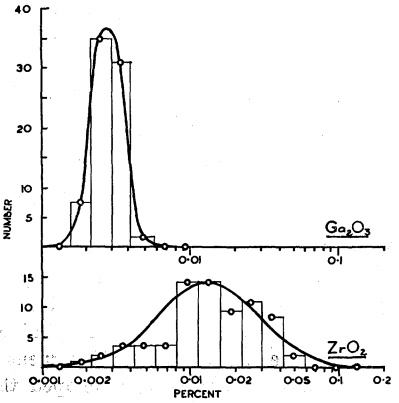

FIG. 1.

based on 75 specimens, which shows this quantitatively for diabase. For comparison, a histogram showing the distribution of zirconium in the same suite of specimens is also given in Fig. 1.

For many of the abundance measurements given during the past two decades, spectrochemical methods were employed. Although these served the desired purpose successfully, for the most part they were semiquantitative, and in many cases the analysis of each element involved a separate operation (re-arcing). In this investigation we have attempted to develop a limited number of general methods, each of which can handle several elements, and each of which may be regarded as a precision method. For example, in one method, Ga, Pb, Cu, and Ag are determined in a single operation; in another, V, Cr, Sc, Y, La, Nd, Zr, Co, Ni, Sr, and Ba are determined. An indication of the reproducibility (precision) is given in Table 1, which shows some replicate analyses of gallium and of zirconium in a specimen of diabase.

TABLE 1

	% Ga_2O_3		% ZrO_2
	0.0027		0.014
	.0027		.014
	.0028		.013
	.0031		.014
	.0028		.012
	.0025		.014
	.0028		.014
	.0027		.013
	.0023		.013
	0.0025		0.012
Mean	0.0027	Mean	0.013

Although a given spectrochemical method may be precise, it may nevertheless introduce a systematic error (bias). The presence of such an error may cause considerable difficulty in correlating sets of analytical data from different laboratories. To reduce possible systematic error to a minimum, all determinations will be calibrated in terms of two naturally occurring standard specimens, one of granite and one of diabase. These specimens have been analyzed spectrochemically and colorimetrically for some elements in several laboratories.

[1] This investigation is part of a general program of spectrographic research carried on in the Department of Geology, MIT, under contract with the Office of Naval Research, Washington, D. C., and under the supervision of H. W. Fairbairn.